

## Mapping of QTLs associated with root and shoot architecture in rice

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### ABSTRACT

A subset of 275 F<sub>7</sub> RIL population derived from a cross between indica cultivars Danteshwari and Dagad Deshi were used to identify the QTLs associated with root and shoot architecture. The phenotypic evaluation was done for eight root traits and four shoot architecture related traits. The RIL population was genotyped using 93 SSR and 11 SNP markers distributed over the entire twelve rice chromosomes. QTL mapping using single marker analysis leads to the identification of 24 markers putatively associated to 10 traits. The total number of markers associated was 1 for root length, 1 for root dry weight, 1 for root/shoot length ratio, 1 for root/shoot dry weight ratio, 3 for root volume, 3 for shoot fresh weight, 3 for tiller number, 6 for shoot dry weight, 8 for shoot length and 11 for total plant length. No markers were associated with root fresh weight and root/shoot fresh weight ratio. Four markers HvSSR01-80, HvSSR01-86, HvSSR01-87 and HvSSR04-26 were found to be strongly associated with total plant height and shoot length. HvSSR05-12 was found to be strongly associated with shoot fresh weight and shoot dry weight. The negative additive effect and LOD > 2.4 of these marker-traits indicating the majority of the favorable alleles for traits are contributed by the landrace, Dagad Deshi which has deep/long roots. The QTLs identified for root and shoot traits will be helpful for the rice molecular breeding programs.

**Key words:** Rice, root, QTL, marker, RIL

Rice is a cereal of high economic and social value, which is used as a staple food by more than half of the world's population. It is the only cereal which is solely produced for human consumption. In the race between increasing world population and food production, the green revolution was principally achieved by increasing crop productivity in irrigated ecosystems. However, success has been limited in increasing rice productivity in rainfed systems, which are prone to frequent drought (Bala Krishna and Satyanarayana, 2013). Drought is a major constraint to rice production in rainfed areas because of unreliable monsoon and uneven distribution of rainfall cause year to year fluctuations in crop yields. Improving drought tolerance to increase productivity and stability has become a challenge. The improved root and shoot traits can contribute to drought tolerance ability of the plant. Maximum root length and root dry weight were good indicators of drought avoidance in

upland rice and plants having deeper root system should colonize a large soil volume and improve the water uptake from the lower layers (Kanbar *et al.*, 2009). The long, thick and extensive root systems specifically contribute highly to drought tolerance (Fukai and Cooper, 1995; Yadav *et al.*, 1997; Comas *et al.*, 2013). It has also been reported that ratios of root weight to shoot weight and root penetration ability were correlated with drought tolerance (Price *et al.*, 1997; Li *et al.*, 2005). Drought is quantitative in nature and therefore, the effect of the individual gene controlling the trait can not be readily identified using conventional method. Molecular markers allow to track the genetic loci linked to such complex traits and help in their indirect selection (Nguyen *et al.*, 1997). Thus, QTL mapping is a powerful tool to understand and improve drought tolerance. Several QTLs have been reported for root and shoot traits in rice (Li *et al.*, 2005; Steele *et al.*,

2006; Komashita *et al.*, 2008; Gomez *et al.*, 2010; Kanagaraj *et al.*, 2010; Zhao *et al.*, 2011; Reddy *et al.*, 2013).

Earlier, Reddy *et al.* (2013) used the same Danteshwari/Dagad deshi RIL population to identify molecular marker for root length and Verma *et al.* (2014a, 2014b) used to identify QTLs for cold tolerance and grain yield. The objectives of this study were to investigate several phe-notypic roots and shoot traits and to identify the underlying QTL in the Danteshwari and Dagad Deshi RIL rice population.

## MATERIALS AND METHODS

Danteshwari is a high yielding popular rice cultivar but moderately susceptible to water stress and Dagad Deshi is a tall deep rooted poor yielder and tolerant to water stress. A cross was made between Danteshwari x Dagad Deshi to develop RIL population through single seed descent method at Research cum Instructional Farm, IGKV, Raipur (Verulkar *et al.*, 2004). Finally two hundred and seventy five F7 recombinant inbred lines were used for QTL mapping.

A set of 275 RILs were evaluated for eight root and four shoot traits in PVC pipe experiment using Randomized Completely Block Design (RCBD) with two replications. Seeds were sown in PVC pipes measuring 1.2 meter in length and 20 cm in diameter, which were filled with a mixture of sandy-clay loam and FYM (Kanbar *et al.*, 2002). After germination, one seedling was allowed to grow in each pipe. Plants were regularly watered and nutrients (micro and macro) were applied in a fixed interval of time. The observations were recorded on five plants for each RIL after 70 days of sowing. Observations consisted of root length, root volume, root fresh weight, root dry weight, shoot length, tiller number, shoot fresh weight and shoot dry weight and were measured as de-scribed previously in rice (Li *et al.*, 2005). For the observation of root and shoot dry weight, plant samples (roots and shoots) were placed in an oven at 65°C for 48 hours. Further, total plant length (shoot length + root length), root/shoot length ratio, root/shoot fresh weight ratio and root/shoot dry weight ratio were computed.

Total 901 SSR markers, including 788 HvSSR (Singh *et al.*, 2010), 108 RM (McCouch *et al.*, 2002) and 5 RGNMS (Parida *et al.*, 2009) were analysed to find markers polymorphic between the parental lines in

4% metaphor agarose gel (Singh *et al.*, 2010). The genotyping of the 275 RILs was carried out with polymorphic SSR markers (Sinha *et al.*, 2015). SNP genotyping was carried with 36 markers (Singh *et al.*, 2010) using Sequenom MALDI-TOF MassArray system as re-ported in Pandit *et al.* (2010).

Data collected from parental lines and RILs were included in the statistical analysis. Descriptive statistics for all traits were calculated for RIL population and Pearson's correlation coeffi-cients between all trait combinations were determined using online software OPSTAT (<http://www.hau.ernet.in>). The genotypic data was used to construct a linkage map using the program MAPMAKER (Lander, 1993). QTL analysis was performed using QTL Cartographer v2.5 software (Zeng, 1994; Wang *et al.*, 2007) to identify the linkage between the marker loci and the phenotypic trait following single marker analysis using mean phenotypic data and genotypic data set derived from RIL population.

## RESULTS AND DISCUSSION

Transgressive segregation was observed for all the twelve traits studied (Fig.1). The transgressive segregants in both the directions indicate that neither parents carried the entire genetic complement of the alleles. The transgressive segregants may be due to complementary action of additive genes that were dispersed among the parents (Yadav *et al.*, 1997; Ali *et al.*, 2000; Kamoshita *et al.*, 2002a). The mean and standard error of RILs were calculated for all phenotypic traits and presented in Table 1.

Pearson's correlation analysis has been adopted to study the correlation between roots and shoot traits. This study revealed root length exhibited significant and positive correlation with root volume, root fresh weight, root dry weight, shoot length, tiller number, shoot fresh weight, shoot dry weight, root/shoot length ratio, total plant length, root/shoot fresh weight ratio and root/shoot dry weight ratio. No significant correlation was observed among root/shoot length, root dry weight, tiller number, shoot fresh weight, shoot dry weight and root/shoot dry weight ratio, suggesting that these traits are sensitive to environment effects.

Since most of the root traits except root/shoot length ratio were found to be positively correlated with shoot length and number of tillers, selection based on the shoot traits is very important, especially because of

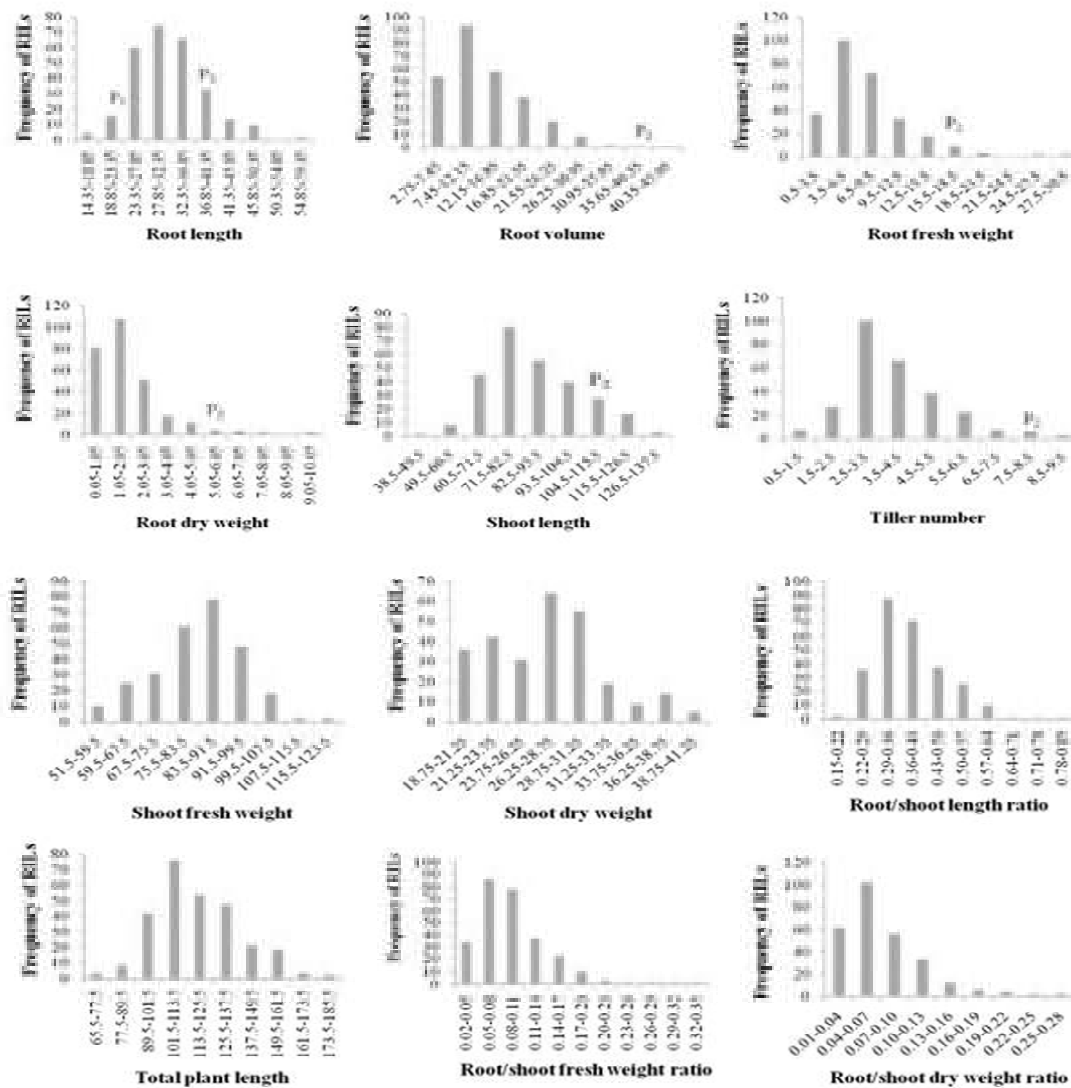


Fig. 1. Frequency distribution of root and shoot traits in RILs derived from Danteshwari x Dagaddeshi

the difficulty in monitoring root development as compared to the shoot. It has also been reported that longer root length, root volume, root fresh weight, root dry weight, root fresh weight/shoot fresh weight ratio and root dry weight/shoot dry weight ratio can be the preferred selection criteria for drought tolerance in rice (Yadav *et al.*, 1997; Hemamalini *et al.*, 2000).

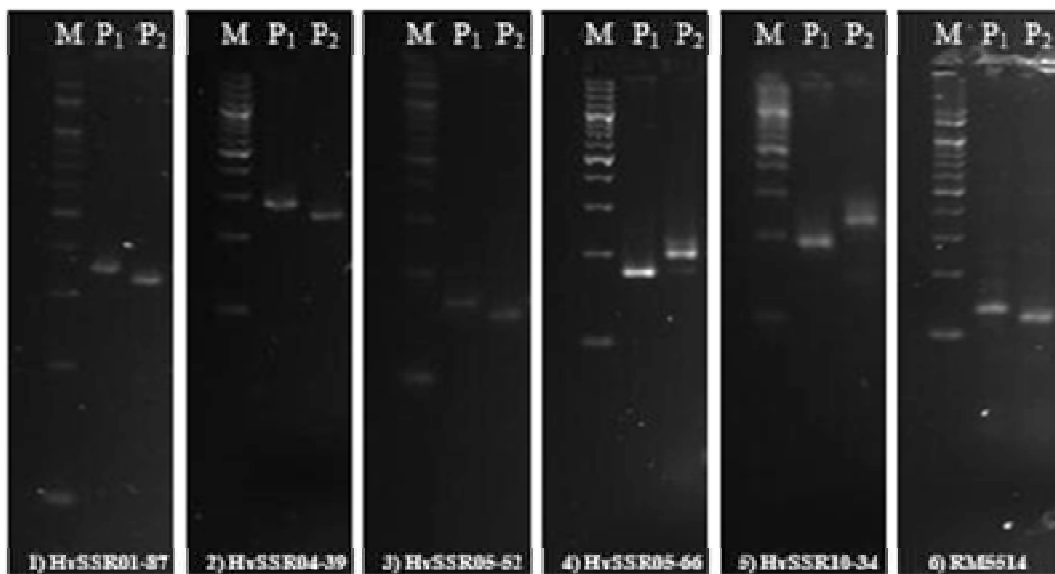
Nine hundred and thirty seven molecular markers were screened for parental polymorphism, 104 (11.9 %) markers (76 HvSSR, 14 RM, 3 RGNMS and 11 SNPs) generated informative polymorphism between the parents (Fig. 2). The primers exhibiting polymorphism on parents were used for PCR

amplification on the 275 RILs (Figure 3).

Low level of parental polymorphism can be attributed to the narrow genetic variation between the parents as both were *indica* ecotypes and adapted to grow in the same rice ecosystem. Several line of evidence are in support which also indicate low level of polymorphism between the parents in the intra-sub-specific (Ali *et al.*, 2000, Subashri *et al.*, 2009; Gomez *et al.*, 2010). The segregation pattern of the 104 polymorphic markers was examined, sixty six markers (63.46 %) deviated significantly from the expected 1:1 ratio ( $\div 2 > 3.84$ ,  $P < 0.05$ ) and exhibited skewed segregation pattern, while 38 (36.54%) markers

**Table 1.** Variation for root and shoot traits in Danteshwari x Dagad Deshi derived F<sub>7</sub> RIL population

Trait	Parents		RILs			
	Danteshwari	Dagad Deshi	Min	Max	Mean	S.D.
Root length (cm)	22.3	41.2	14.35	55.25	31.9	6.47
Root volume (cm <sup>3</sup> )	21	40	3	45	12.98	7.05
Root fresh weight (g)	9.6	18.4	1.44	28.34	7.57	4.45
Root dry weight (g)	2.26	5.3	0.19	9.69	1.86	1.33
Shoot length (cm)	78.4	105.5	39.85	135	86.19	17.08
Tiller number	4	7.5	1	9.25	3.75	1.48
Shoot fresh weight (g)	80.9	108.6	52.4	122.05	83.41	12.64
Shoot dry weight (g)	29.2	40.8	18.75	41.10	27.28	5.01
Root/shoot length ratio	0.28	0.39	0.14	0.78	0.38	0.09
Total plant length	101	146.7	65.5	176.65	118.1	19.37
Root/shoot fresh weight ratio	0.12	0.17	0.02	0.32	0.09	0.05
Root/shoot dry weight ratio	0.08	0.13	0.01	0.26	0.07	0.04



M-100bp ladder

**Fig. 2.** Survey of parental polymorphism on genomic DNA of parents Danteshwari (P<sub>1</sub>)/Dagad Deshi (P<sub>2</sub>)



M-100bp

**Fig.3.** Genotyping on genomic DNA of RILs derived from Danteshwari (P<sub>1</sub>)/Dagad Deshi (P<sub>2</sub>) using SSR marker HvSSR12-51

segregated in the expected ratio. Most of the RILs were skewed towards female parent. Skewed segregation of molecular markers was also observed in other mapping populations (Subashri *et al.*, 2009; Gomez *et al.*, 2010). ). The genetic basis of skewed segregation may be the uneven abortion of male or female gametes or the selective fertilization of particular gametic genotypes (Ali *et al.*, 2000). A framework linkage map was constructed using randomly distributed 104 markers across the 12 rice chromosome covered 2665.5 cM in length with average distance of 25.66 cM between adjacent markers (Figure 4). Low level of polymorphism resulted in poor genome coverage and gaps in the linkage map. Low level of polymorphism and large gaps were also reported in linkage map by Price and Tomos (1997); Ali *et al.* (2000); Amaravathi *et al.* (2008); Geomez *et al.* (2010).

Single marker analysis was performed to estimate association between marker and trait using simple marker function of QTL Cartographer v2.5 software. Single marker analysis identified 24 (18 HvSSR, 4 RM and 2 SNP) markers putatively associated to 10 traits on chromosome 1, 4, 5, 6, 8, 10 and 12 out of 12 investigated at  $P < 0.01$  and  $P < 0.001$  (Table 2, Figure 5). None of the markers associated with root fresh weight and root/shoot fresh weight ratio. The majority of the favorable alleles for traits are contributed by the landrace, Dagad Deshi which has deep/long roots. Deep roots access water from depth and help with no or less leaf rolling and drying. This helps to sustain carbon assimilation and production of biomass under stress. Additive effect of QTLs reveals effect of replacement of a female allele (Danteshwari) by a male (Dagad Deshi) allele.

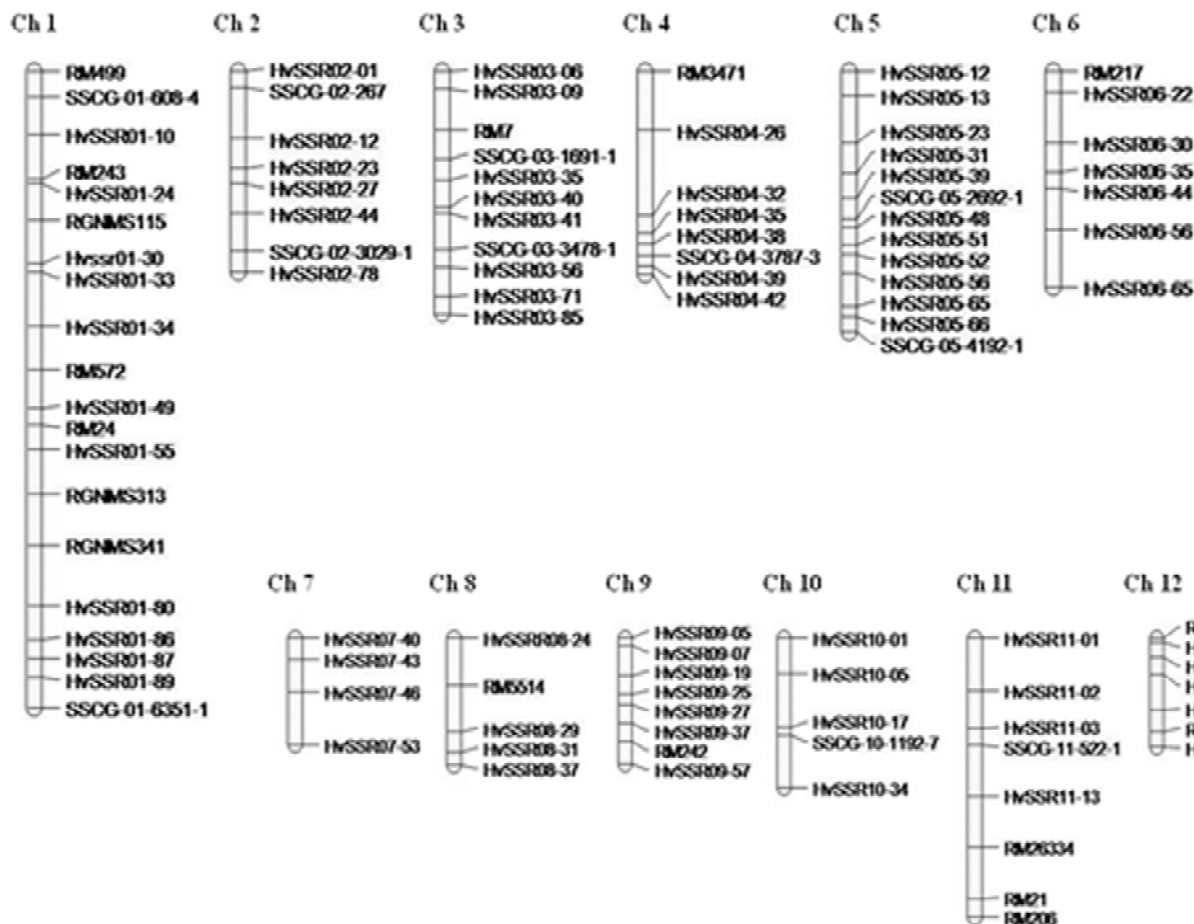


Fig.4. Molecular genetic linkage maps of chromosome generated using 104 polymorphic markers on 275 F7RILs derived from Danteshwari /Dagad Deshi

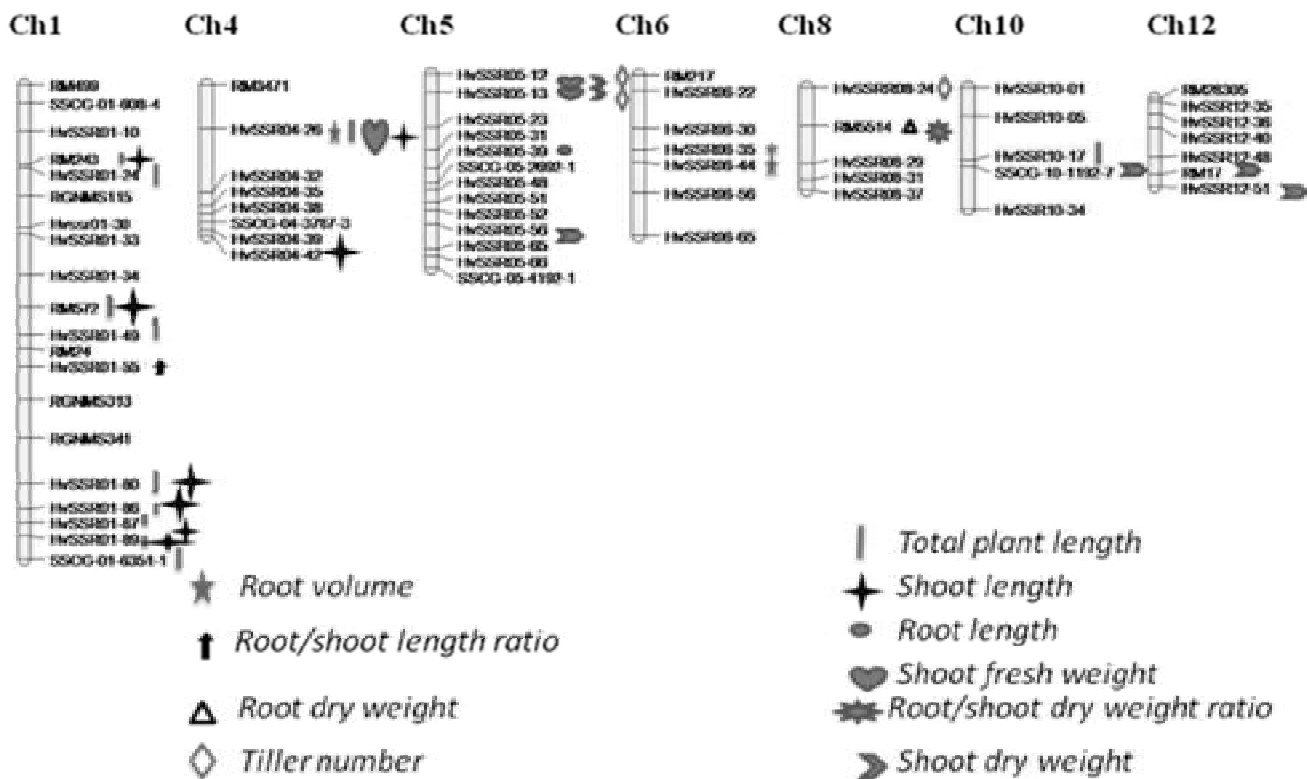
**Table 2.** Marker loci associated with root and shoot traits based on single marker analysis

Trait	Chromosome	Marker	Additive effect	LOD	Probability
Root length	5	HvSSR05-39	-1.23	1.87	0.004**
Root volume	4	HvSSR04-26	-1.38	2.18	0.002**
	6	HvSSR06-35	-1.49	2.17	0.002**
	6	HvSSR06-44	-1.19	1.45	0.01**
Root dry weight	8	RM5514	-0.25	1.87	0.003**
Root/ Shoot length ratio	1	HvSSR01-55	0.02	1.52	0.008**
Root/ Shoot dry weight ratio	8	RM5514	-0.01	1.65	0.006**
Plant height	1	RM243	-3.51	1.69	0.005**
	1	HvSSR01-24	-3.57	1.66	0.006**
	1	RM572	-3.23	1.58	0.007**
	1	HvSSR01-49	-3.59	1.59	0.007**
	1	HvSSR01-80	-4.35	2.6	0.001***
	1	HvSSR01-86	-4.53	2.94	0***
	1	HvSSR01-87	-4.03	2.34	0.001***
	1	HvSSR01-89	-3.2	1.46	0.01**
	1	SSCG-01-6351-1	-3.77	1.67	0.006**
	4	HvSSR04-26	-4.2	2.69	0***
Shoot fresh weight	10	HvSSR10-17	3.25	1.55	0.008**
	4	HvSSR04-26	-2.24	1.79	0.004**
	5	HvSSR05-12	-2.88	2.65	0.001***
Shoot dry weight	5	HvSSR05-13	-2.38	1.96	0.003**
	5	HvSSR05-12	-1.07	2.33	0.001***
	5	HvSSR05-13	-0.98	2.11	0.002**
	5	HvSSR05-56	-0.92	1.74	0.005**
	10	SSCG-10-1192-7	0.88	1.62	0.007**
	12	RM17	0.81	1.5	0.009**
Shoot length	12	HvSSR12-51	0.94	1.81	0.004**
	1	RM243	-2.98	1.57	0.008**
	1	RM572	-2.92	1.66	0.006**
	1	HvSSR01-80	-3.73	2.46	0.001***
	1	HvSSR01-86	-3.94	2.86	0***
	1	HvSSR01-87	-3.87	2.8	0***
	1	HvSSR01-89	-2.84	1.48	0.009**
	4	HvSSR04-26	-3.63	2.58	0.001***
Tiller number	4	HvSSR04-42	-3.23	1.5	0.009**
	5	HvSSR05-12	-0.27	1.65	0.006**
	5	HvSSR05-13	-0.26	1.64	0.006**
	8	HvSSR08-24	-0.26	1.53	0.008**

\*\*P ≤ 0.01, \*\*\* P ≤ 0.001, LOD = Likelihood of odds score

Chromosome 1 had ten loci linked to QTLs for three traits. QTLs identified for shoot length, root/shoot length ratio and total plant length are located near a region flanked by markers HvSSR01-24 and SSCG-01-6351-1. Convergences of QTLs for shoot length, root/shoot length ratio and total plant length at this region showed that this region had genes with high expressivity for cellular elongation, which leads to the crop growth in general. A major gene controlling the semi-dwarf stature *sd-1*, is located near marker RZ730 in Chromosome 1

(Courtois *et al.*, 2000; Kanagaraj *et al.*, 2010). This gene is established to affect many stages of plant morphology and physiology through pleiotropic effects on height, tillering, panicle length, responsiveness to fertilizer, biomass and harvest index (Xia *et al.*, 1991). QTLs associated with root traits that increase yield in upland rice have been investigated (Steele *et al.*, 2013). QTL for shoot length located on chromosome 1 also reported by Diwan *et al.* (2013). Chromosome 4 had two marker loci (HvSSR04-26 and HvSSR04-42)



**Fig.5.** Chromosomes 1,4,5,6,8, 10 & 12 showing putative QTLs associated with root and shoot traits in Danteshwari /Dagad Deshi RILs based on single marker analysis

associated with three traits (root volume, shoot length, shoot fresh weight and total plant length). Lang and Buu (2010) used the Milyang 23/Taiching 189 mapping population and detected regions associated with shoot length on chromosome 4. Four marker loci on Chromosome 5 was associated with root length, tiller number shoot fresh weight and shoot dry weight. Chromosome 6 had two marker loci associated with root volume. Two marker loci on Chromosome 8 were associated with root dry weight, tiller number and root/shoot dry weight ratio. Chromosome 10 had two marker loci associated with shoot dry weight and total plant length. Chromosome 12 had two marker loci associated with shoot dry weight. MacMillan *et al.* (2006a, b) also identified and mapped QTL for basal root thickness, root fresh weight, root dry weight, maximum root length, root/shoot fresh weight ratio, root/shoot dry weight ratio and other root traits.

It was found that some putative QTLs controlling different root and shoot related traits were located in the same chromosome regions or tightly linked together.

Related traits showed common marker association following single marker analysis, HvSSR04-26 (Chromosome 4) for root volume, shoot length, shoot fresh weight and total plant length; RM5514 (Chromosome 8) for root dry weight and root/shoot dry weight ratio; RM243, RM572, HvSSR01-80, HvSSR01-86, HvSSR01-87, HvSSR01-89 (Chromosome 1), HvSSR04-26 (Chromosome 4) for shoot length and total plant length; HvSSR05-12 and HvSSR05-13 (Chromosome 5) for tiller number, shoot fresh weight and shoot dry weight. Developmentally related traits mapping to the same chromosome regions have been reported by Xiao *et al.* (1996); Yadav *et al.* (1997); Hemamalini *et al.* (2000); Li *et al.* (2005); Norton and Price (2009); Gomez *et al.*, (2010) in rice.

In the conclusion, the detection of new putative QTLs associated with root and shoot traits should be useful for rice improvement in the future. The characterization of QTL for root and shoot traits in this study provides insight into the mode of drought tolerance in rice. In addition, the well conserved QTL

associated with drought tolerance and other agronomically important traits can be used as tools to localize syntenic regions in other cereal crops through comparative genomics.

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## REFERENCES

- Ali ML, Pathan MS, Zhang J, Bai, G, Sarkarung S and Nguyen HT 2000. Mapping QTLs for root traits in a recombinant inbred population from two *indica* ecotype in rice. *Theoretical and Applied Genetics* 101: 756-766
- Balakrishna B and Satyanarayana PV 2013. Genetics of brown planthopper (*nilaparvata lugens* L.) Resistance in elite donors of rice (*oryza sativa* L.). *The Bioscan* 8(4): 1413-1416
- Comas LH, Becker SR, Cruz VV, Byrne PF and Dierig DA 2013. Root traits contributing to plant productivity under drought. *Frontiers in Plant Science* 4:1-16
- Courtois B, McLaren G, Sinha PK, Prasad K, Yadav R and Shen L 2000. Mapping QTLs associated with drought avoidance in upland rice. *Molecular Breeding*. 6: 55-66
- Diwan J, Channbyregowda M, Shenoy V, Salimath P and Bhat R 2013. Molecular mapping of early vigour related QTLs in rice. *Research Journal of Biology* 1:24-30
- Fukai S and Cooper M 1995. Development of drought-resistant cultivars using physio-morphological traits in rice. *Field Crops Research*. 40: 67-86
- Gomez SM, Manikanda BN, Satheesh SK, Ramasubramanian T, Chengsong Z, Jeyaprakash P, Senthil A and ChandraBabu R 2010. Molecular mapping and location of QTLs for drought-resistance traits in *indica* rice (*Oryza sativa* L.) lines adapted to target environments. *Acta Physiology Plant* 32: 355-364
- Hemamalini GS, Sashidhar HE and Hittalmani S 2000. Molecular marker assisted tagging of morphological and physiological traits under two contrasting moisture regimes at peak vegetative stage in rice (*Oryza sativa* L.). *Euphytica* 112: 1-10
- Kamoshita A, Babu RC, Manikanda BN and Fukai S 2008. Phenotypic and genotypic analysis of drought-resistance traits for development of rice cultivars adapted to rainfed environments. *Field Crops Research* 109: 1-23
- Kamoshita A, Zhang J, Siopongco J, Sarkarung S, Nguyen HT and Wade LJ 2002a. Effects of phenotyping environment on identification of QTL for rice root morphology under anaerobic conditions. *Crop Science* 42: 255-265
- Kanagaraj P, Prince KSJ, Sheeba JA, Biji KR, Paul SB, Senthil A and Babu RC 2010. Microsatellite markers linked to drought resistance in rice (*Oryza sativa* L.) *Curr. Sci* 98(6): 836-839
- Kanbar A, Shashidhar HE and Hittalmani S 2002. Mapping of QTL associated with root and related traits in population of rice (*Oryza sativa* L.). *Indian J. Genetics* 62(4): 287-290
- Lang NT and Buu BC 2010. Quantitative trait loci influencing drought tolerance in rice (*Oryzasativa* L.). *OmonRice* 17: 22-28
- Kanbar A, Toorchi M and Shashidhar HE 2009. Relationship between yield and morphological characters in rainfed low land rice (*Oryza sativa* L.). *Cereal Research communications* 37(2): 261-268
- Lander ES 1993. MAPMAKER/EXP 3.0 and MAPMAKER/QTL 1.1. Whitehead institute, 9, Cambridge, MA02142, USA.
- Li Z, Mu P, Li C, Zhang H, Li Z, Gao Y and Wang X 2005. QTLmapping of root traits in a doubled haploid population from a cross between upland and lowland japonica rice in three environments. *Theoretical and Applied Genetics* 110: 1244-1252
- MacMillan K, Emrich K, Piepho HP, Mullins CE and Price AH 2006a. Assessing the importance of genotype by environment interaction for root traits in rice using a mapping population II: conventional QTL analysis. *Theoretical and Applied Genetics* 113: 953-964
- MacMillan K, Emrich K, Piepho HP, Mullins CE and Price AH 2006b. Assessing the importance of genotype by environment interaction for root traits in rice using a mapping population I: a soil-filled box screen. *Theoretical and Applied Genetics* 113: 977-986
- McCouch SR, Teytelman L, Xu Y, Lobos KB, Clare K, Walton M, Fu B, Maghirang R, Li Z, Xing Y, Zhang Q, Kono I, Yano M, Fjellstrom R, DeClerck G, Schneider D, Cartinhour S, Ware D and Stein L 2002. Development and mapping of 2240 new SSR markers for rice (*Oryza sativa* L.). *DNA Research* 9: 199-207
- Nguyen HT, Babu RC and Blum A 1997. Breeding for drought



- tolerance in rice: physiology and molecular genetics considerations. *Crop Science* 37: 1426-1434
- Norton GJ and Price AH 2009. Mapping of quantitative trait loci for seminal root morphology and gravitropic response in rice. *Euphytica* 166(2): 229-237
- Pandit A, Rai V, Bal S, Sinha S, Kumar V, Chauhan M, Gautam RK, Singh R, Sharma PC, Singh AK, Gaikwad K, Sharma TR, Mohapatra T and Singh NK 2010. Combining QTL mapping and transcriptome profiling of bulked RILs for identification of functional polymorphism for salt tolerance genes in rice (*Oryza sativa* L.). *Mol Genet Genomics* 284:121-136
- Parida SK, Dalal V, Singh AK, Singh NK and Mohapatra T 2009. Genic non-coding microsatellites in the rice genome characterization, marker design and use in assessing genetic and evolutionary relationships among domesticated groups. *BMC Genomics*. 10: 140
- Price AH. and Tomos AD 1997. Genetic dissection of root growth in rice. II Mapping quantitative trait loci using molecular markers. *Theoretical and Applied Genetics* 95: 143-152
- Reddy ES, Verma SK, Xalxo SM, Saxena RR and Verulkar SB 2013. Identification of molecular marker(s) for root length in rice (*oryza sativa* l.). *The Bioscan* 8(4): 1511-1514
- Redona ED and Mackill DJ 1996b. Mapping quantitative trait loci for seeding vigor using RFLPs. *Theoretical and Applied Genetics* 92: 395-402
- Singh A, Singh PK, Singh R, Pandit A, Mahato AK, Gupta DK, Tyagi K, Singh AK, Singh NK and Sharma TR 2010. SNP haplotypes of the BADH1 gene and their association with aroma in rice (*Oryza sativa* L.). *Molecular Breeding* 26: 325-338
- Singh H, Deshmukh RK, Singh A, Gaikwad K, Sharma, TR, Mohapatra T and Singh NK 2010. Highly variable SSR markers suitable for rice genotyping using agarose gels. *Molecular Breeding* 25: 359-364
- Sinha S, Verulkar, SB and Kotasthane AS. 2015. Identification of QTLs for root and shoot traits in RIL population of rice (*Oryza sativa* L.). *Indian J. Genet* 75(1): 49-56
- Steele KA, Price AH, Shashidhar HE and Witcombe JR 2006. Marker assisted selection to introgress rice QTLs controlling root traits into an Indian upland rice variety. *Theoretical and Applied Genetics* 112: 208-221
- Steele KA, Price AH, Witcombe JR, Shrestha R, Singh BN, Gibbons JM and Virk DS 2013. QTLs associated with root traits increase yield in upland rice when transferred through marker assisted selection. *Theoretical and Applied Genetics* 126: 101-108
- Subashri M, Robin S, Vinod, KK, Rajeswari S, Mohanasundaram K and Raveendran TS 2009. Trait identification and QTL validation for reproductive stage drought resistance in rice using selective genotyping of near flowering RILs. *Euphytica* 166: 291-305
- Verma SK, Saxena RR, Saxena RR, Xalxo MS and Verulkar SB 2014b. QTL for grain yield under water stress and non-stress conditions over years in rice (*Oryza sativa* L.). *Australian J. Crop Sci* 8(6): 916-926
- Verma SK, Xalxo MS, Saxena RR and Verulkar SB 2014a. Identification of QTLs for cold tolerance at seedling stage in rice (*Oryza sativa* L.). *Indian J. Genet* 74(1): 86-89
- Verulkar SB, Khillare A, Raisagar A, Dudhare MS and Shrivastava MN 2004. Progress in developing drought tolerant rice cultivars for eastern India. In *Resilient crops for water limited environments: Proceedings of a workshop held at Mexico*. pp. 243-244
- Wang S, Basten CJ and Zeng ZB 2007. *Windows QTL cartographer 2.5*
- Xia BS, Hanada K and Kizuzhi F. 1991. Character expression of the semi-dwarfism gene *sd1* in rice. Effect of nitrogen levels on the expression of some agronomic characteristics. *Jpn. J. Crop Science* 60: 36-41.
- Xiao J, Li J, Yuan L and Tanksley SD 1996. Identification of QTLs affecting traits of agronomic importance in recombinant inbred population derived from a subspecific cross. *Theoretical and Applied Genetics* 92: 230-244
- Yadav R, Courtois B, Huang N and McLaren G 1997. Mapping genes controlling root morphology and root distribution on a double-haploid population of rice. *Theoretical and Applied Genetics* 94: 619-632
- Zeng ZB 1994. Precision mapping of quantitative trait loci. *Genetics* 136: 1457-1468
- Zhao K, Tung CW, Eizenga GC, Wright MH, Ali ML, Price AH, Norton GJ, Islam MR, Reynolds A, Mezey J, McClung AM, Bustamante CD and McCouch SR. 2011. Genome-wide association mapping reveals a rich genetic architecture of complex traits in *Oryza sativa*. *Nat. Commun* 2: 467